

U.S. Application No.: 10/502,132
AMENDMENT B

Attorney Docket: 3968-120

REMARKS

Review and reconsideration of the Office Action of December 13, 2005, is respectfully requested in view of the above amendments and the following remarks.

No new matter has been added to the claims or the specification.

For the reasons set forth below, Applicants believe that all the claims are now in conditions for allowance.

Status of the Claims

Claims 1-19 were pending.

Claim 6 was withdrawn from further consideration pursuant to 37 C.F.R. §1.142(b) as being drawn to a non-elected invention, there being no allowable generic or linking claim.

Applicants hereby cancel claim 6.

New claim 20 finds support in, e.g., paragraph [00126], line 3, of the specification as filed.

New claim 21 finds support in paragraph [00095], line 2, and paragraph [00100], line 2, and paragraph [00116], line 2, of the specification as filed.

New claim 22 claims the range defined by the endpoints of claims 20 and 21.

Claims 1-5, 7-19 are rejected and are examined herein.

Office Action

Turning now to the Office Action in greater detail, the paragraphing of the Examiner is adopted.

Claim Rejections -- 35 U.S.C. §112

Examiner finds claims 4 and 7 unclear as to whether one group or all groups are included in the claimed composition.

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In response, Applicants submit that the term "or" signaled that only one of the recited groups need be present in the claimed composition. However, for greater clarity, Applicants now use the "at least one of ... and ..." form.

Claim 8 is rejected for omission of one end of the range of carbon atoms.

In response, Claim 8 is amended, with support found, e.g., in claim 7.

Claim 10 is rejected – the meaning of the term "lacuna" is not clear.

Applicants delete this term from the claim.

Withdrawal of the rejections is respectfully requested.

IDS

Applicants appreciate the Examiner's initialing of form PTO 1449 filed with the IDS, indicating that all references have been considered.

Claim Rejections – 35 U.S.C. §103

Claims 1-5,7-19 are rejected under 35 U.S.C. §103 (a) as being obvious over Clarkson et al. (U.S. Patent Application 2001/0036964 A1) in view of Eggensperger et al. (U.S. Patent 5,670,160) and further in view of Riebel et al. (U.S. Patent Application 2003/0100613A1).

Clarkson is cited for teaching an anti-microbial composition comprising an iron (III) chelator, water, and a polyhydric alcohol. The Examiner admits that Clarkson fails to teach particular combinations of alkyl diols or specific preservatives (as listed in Applicants claim 12).

Applicants respectfully traverse.

Clarkson is merely illustrative of the state of the art prior to the present invention.

As discussed in the background section of the present specification, in the cosmetic and pharmaceutical industry and also in the food industry there is an ongoing need for agents with antimicrobial properties, in particular for the preservation of perishable products and for the direct cosmetic or therapeutic treatment of microorganisms and particularly those that can give rise to body odor (including underarm and foot odor), acne, mycoses or the like. The search for

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suitable (active) is difficult because there is no clear dependence between the chemical structure of a substance, on the one hand, and its biological activity towards specific microorganisms (germs) and its stability, on the other hand. Furthermore, there is no predictable relationship between the antimicrobial action, toxicological acceptability, tolerance by the skin and the stability of a substance.

The present invention is based on the surprising finding that straight-chain 1,2-alkanediols with a chain length in the range of 5 to 10 C atoms exhibit a synergistically intensified antimicrobial effect, at least against selected germs, if they are combined with a second or further straight-chain 1,2-alkanediols with different chain lengths in the same range.

In particular, it has been found that the mixtures according to the invention of two, three or more straight-chain 1,2-alkanediols of different chain lengths are outstandingly suitable for use for the preservation of articles that would otherwise be perishable.

As discussed in the background section of the specification, and as exemplified by the numerous references discussed therein, experts in the field had already concerned themselves extensively with the antimicrobial properties of 1,2-diols. However, hitherto there has been no indication that mixtures of two, three or more straight-chain 1,2-alkanediols, the chain lengths of which (i) are different and (ii) in each case are in the range of 5 to 10 C atoms, possess an antimicrobial action (at least against selected germs) that is distinctly improved in the individual case. The prior art also gave no incentive to use such mixtures (combinations) as antimicrobial active compounds.

In fact, considering the comprehensive research of record on the antimicrobial activity of individual diols having a chain length in the range of 5 to 10 C atoms, it must be seen as particularly surprising that mixtures of two, three or more straight-chain 1,2-alkanediols, the chain lengths of which (i) are different and (ii) in each case are in the range of 5 to 10 C atoms display a strongly synergistic activity and are clearly superior to the individually dosed 1,2-diols having chain lengths in the same range in the same concentration, in particular with regard to the reduction in germ time. In particular, a CFU value (CFU = number of colony-forming

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units) of 0 can be achieved in the individual case only with the said mixtures according to the invention.

The mixtures according to the invention develop their synergistic action against a multiplicity of Gram-positive bacteria, Gram-negative bacteria, moulds and yeasts. There is a particularly good action against Gram-negative bacteria such as *Escherichia coli* and *Pseudomonas aeruginosa*, against yeasts such as *Candida albicans* and against fungi such as *Aspergillus niger*. In this context the very good activity of the 1,2-diol mixtures according to the invention against *Aspergillus niger*, a mould that can be controlled only with very great difficulty, is to be regarded as particularly advantageous, since Applicant's own studies have shown that when individual 1,2-diols with a chain length in the range of 5 to 10 C atoms are used the CFU value thereof cannot be reduced to the value 0.

The present invention also relates to corresponding methods for the cosmetic and/or therapeutic treatment of germs and specifically, in particular, of (a) microorganisms causing body odour, (b) microorganisms causing acne and/or (c) microorganisms causing mycoses, comprising the topical application of the claimed antimicrobially effective amount of a mixture of two, three or more straight-chain 1,2-alkanediols, the chain lengths of which (i) are different and (ii) in each case are in the range of 5 to 10 C atoms, the proportions of the said diols in the mixture being set such that their antimicrobial action is synergistically intensified.

As Applicant's own research now showed, the synergistically active mixtures according to the invention consisting of two, three or more straight-chain 1,2-alkanediols, the chain lengths of which (i) are different and (ii) in each case are in the range of 5 to 10 C atoms, not only have a good action against the germs already mentioned above but also against *Staphylococcus epidermidis*, *Brevibacterium epidermidis*, *Propionibacterium acnes* as well as against *Trichophyton* and *Epidermophyton* species, so that they can also be used as agents for the treatment (control) of underarm odour and foot odour and of body odour in general, as agents for the control of acne, as anti-dandruff agents and for the treatment of mycoses (in particular dermatomycoses (sic)) (again see Table 1).

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Table 1:

Microorganisms:	
<i>Staphylococcus epidermidis</i>	underarm odour, body odour in general
<i>Staphylococcus aureus</i>	atopic eczemas; wound infection
<i>Corynebacterium xerosis</i>	underarm odour
<i>Brevibacterium epidermidis</i>	underarm odour; foot odour
<i>Propionibacterium acnes</i>	acne
<i>Escherichia coli</i>	wound infection
<i>Pseudomonas aeruginosa</i>	wound infection
<i>Malassezia furfur</i> (syn. <i>Pityrosporum ovale</i>)	development of dandruff
<i>Candida albicans</i>	general candidoses
<i>Trichophyton mentagrophytes</i>	skin and nail mycoses
<i>Trichophyton rubrum</i>	skin and nail mycoses
<i>Epidermophyton floccosum</i>	skin and nail mycoses
<i>Aspergillus niger</i>	mould infestation

A microorganism that causes acne is *Propionibacterium acnes*, which is a germ that grows anaerobically. The cosmetics industry is continually looking for agents for the treatment of this germ and other microorganisms that cause acne.

All areas of the human skin can be infested by mycoses (in particular dermatomycoses and nail mycoses). Areas of the skin on which moisture and warmth can build up as a result of wearing clothing, shoes or jewellery are particularly frequently affected. Fungus diseases of the fingernail and toenail regions are experienced as being particularly unpleasant. Various species of *Trichophyton* and *Epidermophyton* frequently have decisive responsibility for the formation of mycoses. The cosmetics industry is continuously searching for novel agents for the treatment of microorganisms causing these and other mycoses.

The present specification provides extensive evidence of the unexpected strong synergism found with the claimed combinations.

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Example 1, discussed beginning at page 29 of the specification, tested preservation by synergistically active 1,2-diol-mixtures against *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Candida albicans*, and *Aspergillus niger*. The results of the preservative loading tests for the 1,2-diols and 1,2-diol mixtures tested are given in Table 3. Surprisingly, it was found that 1,2-diol mixtures consisting of defined mass ratios of 1,2-pentanediol, 1,2-hexanediol, 1,2-octanediol and/or 1,2-decanediol have a far greater activity than the individual substances metered in the same concentration. This is shown, in particular, in the residual germ counts remaining after 28 days. Binary and ternary mixtures of 1,2-hexanediol with 1,2-pentanediol and/or 1,2-octanediol proved to be particularly effective here. In the case of all 5 test germs (*Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Candida albicans* and *Aspergillus niger*) it was possible to reduce the concentration of colony forming units (CFU) to the preferred target 0 value within the chosen time period.

As can be seen from Table 3 and Fig. 1 (test for adequate preservation for *Aspergillus niger* over a period of 28 days; logarithmic plot of the reduction in the germ count for 1,2-hexanediol (3% in O/W emulsion), for 1,2-octanediol (3%) and for a 1,2-hexanediol/1,2-octanediol mixture (mass ratio 2:1; dosage likewise 3%), it was possible in the case of *Aspergillus niger*, a germ that is particularly problematical with regard to the preservation of industrial products, to reduce the germ count to 0 within 28 days by using the mixtures according to the invention. On the other hand, the individual substances (1,2-pentanediol, 1,2-hexanediol, 1,2-octanediol and 1,2-decanediol; Table 3) tested for comparison purposes, likewise in a dosage of 3%, did not enable a reduction in the number of colony forming units (CFU) to the desired 0 value in the case of *Aspergillus niger*. The test series (Fig. 1 and Tab. 3) thus show, by way of example, that 1,2-diol mixtures consisting of at least two different members of the group that consists of 1,2-pentanediol, 1,2-hexanediol, 1,2-octanediol and 1,2-decanediol have a synergistically intensified activity.

Based on the available data, the synergistic intensification of the activity of the diol mixtures according to the invention can also be demonstrated on the basis of the Kull equation

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(F.C. Kull et al.; Applied Microbiology Vol. 9, p. 538-541 (1961); David C. Steinberg; Cosmetics & Toiletries Vol. 115 (No. 11), p. 59-62; November 2000; see also Table 4 for the method of calculation). The Kull equation enables the pure substances and the active compound mixtures prepared therefrom to be compared in respect of their antimicrobial activity. With this equation the so-called synergy index (SI), which is a measure for a synergistic activity, but also for a possible antagonistic activity, of a mixture having an antimicrobial action, is determined. A synergistic effect is evident if the SI value determined is less than 1. On the other hand, if an SI of precisely 1 is calculated, there is a pure additive effect of two substances having an antimicrobial action. In the case of an SI value greater than 1, on the other hand, there is a (frequently undesired) antagonistic effect.

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Table 4: Calculation of the synergy index (SI) for 1,2-hexanediol/1,2-octanediol (Mass ratio: 2:1; dosage in O/W emulsion: 3%; test germ: *Aspergillus niger*)

	A	B	C
	1,2-hexanediol	1,2-octanediol	1,2-hexanediol + 1,2-octanediol; Mass ratio: 2:1
<i>Aspergillus niger</i> : 14 days [CFU/ml]	28000	1000	300
Kull Equation: $SI = CxD/A + CxE/B$			
A: Germ count for substance A	28000		
B: Germ count for substance B	1000		
C: Germ count for mixture of A + B	300		
D: Amount of A in C	0.66		
E: Amount of B in C	0.33		
SI: Synergy Index	0.106		
Literature : Synergy Index:			
D.C.Steinberg; Cosmetics & Toiletries 115 (11); p. 59-62 (2000)			
F.C.Kull et al.; Applied Microbiology 9; p. 538-541 (1961)			

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Example 2 showed the determination of the minimum inhibitory concentrations for various germs which, for example, are responsible for body odour goes back to test series in which the particularly relevant germs *Staphylococcus epidermidis*, *Corynebacterium xerosis* and *Brevibacterium epidermidis* were tested. In addition to the MIC values for *Staphylococcus epidermidis*, *Corynebacterium xerosis* and *Brevibacterium epidermidis*, the corresponding synergy indices of the synergistically active mixtures according to the invention were determined in these test series (cf. Table 7). In addition, the MIC determinations showed that the 1,2-diol mixtures claimed also have a surprisingly good action against further test germs such as *Trichophyton mentagrophytes*, *Epidermophyton floccosum*, *Propionibacterium acnes*, as a result of which the diol mixtures claimed can also be used as agents against mycoses or acne.

The MIC values for 1,2-hexanediol, 1,2-octanediol and for a mixture of the two diols in a mass ratio of 2 parts of hexanediol to 1 part of octanediol were determined in accordance with the general test conditions described (cf. Table 7).

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Table 7:

MIC values [ppm] for 1,2-hexanediol, 1,2-octanediol and for a C6/C8 diol mixture (2:1)					
Determination of the Synergy Indices (SI) in accordance with the Kull et al. ^{1,2} equation					
Microorganism	Strain No.	MIC C6	MIC C8	MIC C6/C8 2:1	SI: C6/C8; 2:1
<i>Staphylococcus epidermidis</i>	ATCC 12228	25000	12500	6250	0.55
<i>Corynebacterium xerosis</i>	ATCC 7711	12500	6250	6250	0.66
<i>Brevibacterium epidermidis</i>	ATCC 35514	25000	3125	6250	0.83
<i>Propionibacterium acnes</i>	ATCC 11829	25000	6250	3125	0.25
<i>Malassezia furfur</i>	DSM 6171	12500	50000	50000	2.97
<i>Trichophyton mentagrophytes</i>	CBS 26379	6300	1562	1562	0.49
<i>Epidermophyton floccosum</i>	CBS 55384	6250	3125	1562	0.32
¹ F.C.Kull et al.; Applied Microbiology 9; p. 538-541 (1961)					
² D.C.Steinberg; Cosmetics & Toiletries 115 (11); p. 59-62 (2000)					

The results shown in Table 7 show, for 1,2-hexanediol and 1,2-octanediol by way of example, the synergistic intensification of the activity of the 2:1 mixture of the two diols. Accordingly, microorganisms such as *Staphylococcus epidermidis*, *Brevibacterium epidermidis*,

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Corynebacterium xerosis, *Propionibacterium acnes*, *Trichophyton mentagrophytes* and *Epidermophyton floccosum* are also clearly more strongly inhibited by the 1,2-diol mixture in direct comparison with the corresponding individual substances. The synergy indices determined on the basis of the MIC values with the aid of the Kull equation are also shown in Table 7. The SI values clearly show that the 1,2-hexanediol/1,2-octanediol diol mixture has a synergistically intensified activity and, in addition to its excellent activity as a preservative (cf. Example 1) can also preferentially be used for controlling body odour (SI *Staphylococcus epidermidis*: 0.55; SI *Corynebacterium xerosis*: 0.66; SI *Brevibacterium epidermidis*: 0.83), for controlling acne (SI *Propionibacterium acnes*: 0.25) and for controlling the skin and nail mycoses caused by *Trichophyton* and *Epidermophyton* species (SI *Trichophyton mentagrophytes*: 0.49; SI *Epidermophyton floccosum*: 0.32). On the other hand, in the case of *Malassezia furfur* it was not possible to demonstrate a synergistic effect for the 1,2-hexanediol/1,2-octanediol mixture (SI *Malassezia furfur*: 2.97, i.e. antagonistic effect).

Further yet, Example 3 demonstrated preservation by mixtures of (a) synergistic mixtures of 1,2-alkanediols with (b) further preservatives

The results of the preservative loading tests for the combinations of active compounds tested, consisting of a synergistic mixture of 1,2-alkanediols according to the invention and a further preservative, are given in Tables 9 and 10 for the system 1,2-hexanediol/1,2-octanediol/Euxyl K400 by way of example. Surprisingly, it was found that not only 1,2-diol mixtures themselves but also combinations of 1,2-diol mixtures with further preservatives are able to achieve a significant synergistic intensification of activity compared with the individual substances metered in the same concentration. In the example mentioned, this is shown in particular in the residual germ counts for *Aspergillus niger* remaining after 28 days. As can be seen from Table 9, in the case of *Aspergillus niger*, a germ that is particularly problematical with regard to the preservation of industrial products, it was possible to reduce the germ count to 2,800 within 28 days by using formulation C. On the other hand, the diol mixture according to formulation B (1,2-hexanediol + 1,2-octanediol; mass ratio 1:1) tested in a dosage

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of 1% and formulation A (containing Euxyl K400), which was also tested in a concentration of 0.1% for comparison purposes, did not enable such a significant reduction in the number of colony forming units (CFU) in the case of *Aspergillus niger*. The test series (Tab. 9) thus shows, by way of example, that mixtures of active compounds consisting of (a) at least two different members of the group that consists of 1,2-pentanediol, 1,2-hexanediol, 1,2-octanediol and 1,2-decanediol and (b) a further preservative can possess a synergistic, further improved action.

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Table 9: Testing for adequate preservation for a combination of active compounds consisting of a synergistically active diol mixture according to the invention (1,2-hexanediol and 1,2-octanediol) and a further preservative Euxyl K400; [CFU/ml]

1. O/W emulsion with formulation A (0.1% Euxyl K400)					
Days	Escherichia Coli	Pseudomonas aeruginosa	Staphylococcus Aureus	Candida albicans	Aspergillus niger
0	300,000	320,000	310,000	310,000	280,000
1	200	0	0	<100	220,000
2	<100	0	0	<100	140,000
4	0	0	0	0	120,000
7	0	0	0	0	200,000
14	0	0	0	0	160,000
28	0	0	0	0	32,000
2. O/W emulsion with formulation B (0.5% hexanediol + 0.5% octanediol)					
Days	Escherichia coli	Pseudomonas aeruginosa	Staphylococcus Aureus	Candida albicans	Aspergillus niger
0	300,000	320,000	310,000	310,000	280,000
1	0	0	0	3,200	180,000
2	0	0	0	200	160,000
4	0	0	0	100	140,000
7	0	0	0	0	180,000
14	0	0	0	0	120,000
28	0	0	0	0	60,000
3. O/W emulsion with formulation C (0.25% hexanediol + 0.25% Octanediol + 0.05% Euxyl K 400)					

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Days	<i>Escherichia coli</i>	<i>Pseudomonas aeruginosa</i>	<i>Staphylococcus Aureus</i>	<i>Candida albicans</i>	<i>Aspergillus niger</i>
0	300,000	320,000	310,000	310,000	280,000
1	<100	<100	<100	<100	20,000
2	0	0	0	0	12,000
4	0	0	0	0	10,000
7	0	0	0	0	8,000
14	0	0	0	0	8,000
28	0	0	0	0	2,800

Based on the available data, the synergistic intensification of the activity of the combination of active compounds consisting of (a) a synergistically active mixture of 1,2-alkanediols according to the invention (for example 1,2-hexanediol and 1,2-octanediol) and (b) a further preservative (for example Euxyl K400) can also be demonstrated on the basis of the Kull equation (F.C. Kull et al.; Applied Microbiology Vol. 9, p. 538-541 (1961); David C. Steinberg; Cosmetics & Toiletries Vol. 115 (No. 11), p. 59-62; November 2000; see also Table 10 for the method of calculation). The Kull equation enables the pure substances and the active compound mixtures prepared therefrom to be compared in respect of their antimicrobial activity. With this equation the so-called synergy index (SI), which is a measure for a synergistic activity, but also for a possible antagonistic activity, of a mixture having an antimicrobial action, is determined. A synergistic effect is evident if the SI value determined is less than 1. On the other hand, if an SI of precisely 1 is calculated, there is a pure additive effect of two substances having an antimicrobial action. In the case of an SI value greater than 1, on the other hand, there is a (frequently undesired) antagonistic effect.

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Table 10: Calculation of the synergy index (SI) of a 1,2-hexanediol/1,2-octanediol/Euxyl K400 mixture (Formulation C) consisting of equal parts of the comparison 1,2-alkanediol mixture (Formulation A) and of the comparison solution Euxyl K400 (Formulation B) (mass ratio 1:1.; dosage in O/W emulsio: test germ: *Aspergillus niger*)

	A	B	C
	1,2-hexanediol 0.5% + 1,2-octanediol 0.5%	Euxyl K400 0.1%	1,2-hexanediol (0.25%) + 1,2-octanediol (0.25) + EuxylK400 (0.05%);
<i>Aspergillus niger</i> : 28 days [CFU/ml]	60000	32000	2800
Kull equation: $SI = CxD/A + CxE/B$			
A: Germ count for substance A	60000		
B: Germ count for substance B	32000		
C: Germ count for mixture of A + B	2800		
D: Amount of A in C	0.5		
E: Amount of B in C	0.5		
SI: Synergy Index	0.066		
Literature : Synergy Index:			
D.C.Steinberg; Cosmetics & Toiletries 115 (11); p. 59-62 (2000)			
F.C.Kull et al.; Applied Microbiology 9; p. 538-541 (1961)			

Accordingly, Applicants have demonstrated the surprising synergistic enhanced antimicrobial activity commensurate in scope with the claims. This synergism is not suggested

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in Clarkson. The Examiner is also requested to note that there no clear dependence between the chemical structure of a substance, on the one hand, and its biological activity towards specific microorganisms (germs) and its stability, on the other hand. Furthermore, there is no predictable relationship between the antimicrobial action, toxicological acceptability, tolerance by the skin and the stability of a substance. This further supports the unexpected nature of the surprising finding that straight-chain 1,2-alkanediols with a chain length in the range of 5 to 10 C atoms exhibit a synergistically intensified antimicrobial effect, at least against selected germs, if they are combined with a second or further straight-chain 1,2-alkanediols with different chain lengths in the same range.

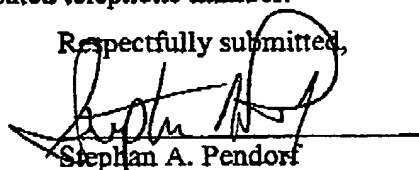
Turning finally to the secondary references, Eggensperger et al and Riebel et al are not cited for teaching combinations of straight-chain 1,2-alkanediols with a chain length in the range of 5 to 10 C atoms, but rather merely the use of supplemental antimicrobial agents.

Accordingly, it is respectfully submitted that the claims as amended are in condition for allowance.

The Commissioner is hereby authorized to charge any additional fees which may be required at any time during the prosecution of this application without specific authorization, or credit any overpayment, to Deposit Account Number 50-0951.

Favorable consideration and early issuance of the Notice of Allowance are respectfully requested. Should further issues remain prior to allowance, the Examiner is respectfully requested to contact the undersigned at the indicated telephone number.

Respectfully submitted,



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